Carbon Isotope Discrimination in Orchardgrass and Ryegrasses at Four Irrigation Levels

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ABSTRACT

Availability of irrigation water is often the limiting factor in the establishment of improved pastures in the U.S. Intermountain Region, and grasses used for pasture should have enhanced water-use efficiency (WUE). Objectives of this study were to describe (i) relationships between carbon isotope discrimination (Δ) and dry matter (DM) yield, and (ii) trends in Δ across four water levels (WL 2, wettest, to WL 5, driest) and two harvest dates under frequent defoliation in cultivars of orchardgrass (*Dactylis glomerata* L.) perennial ryegrass, (*Lolium perenne* L.), intermediate ryegrass (*L. ×hybridum* Hausskn), and festulolium [×*festulolium braunii* (K. Richt.) A. Camus.]. Within all WLs, orchardgrass cultivars exhibited higher WUE than the ryegrasses, which includes intermediate ryegrass and festulolium. Significantly more variation for Δ within WLs was observed in the ryegrasses than orchardgrass. For orchardgrass, DM yield and Δ were not correlated at WLs 2 to 4. However, they were negatively correlated at WL 5. Correlations between Δ and DM yield were not significant at WLs 2 and 3 for the ryegrasses, but were positive at WLs 4 and 5. To avoid possible declines in DM yield in breeding programs to increase WUE in orchardgrass and the ryegrasses, DM yield should also be monitored.

Cool-season grass production in the Intermountain Region of the western USA is closely associated with available soil water. Consequently, plant materials for these areas should have adaptations that allow them to use limited water efficiently (Barker et al., 1989; Johnson et al., 1990; Asay et al., 1998). Martin and Thorstenson (1988) suggested that efficient water use by plants should allow plants to extend water availability. Historically, germplasm improvement in orchardgrass and forage-type perennial ryegrass has focused on forage yield and quality, disease resistance, and adaptation to temperate areas (Stratton and Ohm, 1989; Balasko et al., 1995; Christie and McElroy, 1995; Jung et al., 1996; Casler et al., 1997).

Improved WUE may provide a means to increase forage production in water-limited environments (Asay and Johnson, 1983; Barker et al., 1989). Direct evaluation of WUE, which involves precise measurements of individual plant growth and water consumption, are usually not feasible in field breeding nurseries and makes selection for WUE extremely difficult (Frank et al., 1987; Barker et al., 1989).

Carbon isotope discrimination has been proposed as a criterion to select superior genotypes with improved WUE in C_3 crop species (Farquhar et al., 1982; Farquhar and Richards, 1984; Johnson et al., 1990; Hall et al., 1994). This indirect method of evaluating WUE involves analysis of stable carbon isotope composition (C^{12} and C^{13}) of plant tissues (Johnson and Rumbaugh, 1995). A negative relationship between Δ and WUE has been reported in wheat, *Triticum aestivum* L. (Farquhar and Richards, 1984; Condon et al., 1987); peanut, *Arachis hypogaea* L. (Hubick et al., 1988; Wright et al., 1988); orchardgrass, tall fescue (*Festuca arundinacea* Schreb.), and perennial ryegrass (Johnson and Bassett, 1991); alfalfa, *Medicago sativa* L. (Johnson and Tieszen, 1994); crested wheatgrass [*Agropyron desertorum* (Fisch. ex Link) Shult.] (Johnson et al., 1990; Read et al., 1991); and Altai wildrye, *Leymus angustus* (Trin.) Pilg. (Johnson et al., 1990). Farquhar et al. (1988) attributed this negative association between WUE and Δ to independent linkages through internal CO_2 concentration inside the leaf.

The line-source irrigation system is a unique method that was developed to evaluate plant growth under a gradient of WLs (Hanks et al., 1976). This system produces a nearly linear water application gradient with the amount of irrigation declining with distance from the sprinkler line. The major limitation with the line-source sprinkler system is that WLs for each plot cannot be randomly assigned. Consequently, a valid statistical test for significance of the main effects due to WL can not be made (Hanks et al., 1980). However, tests for other effects and their interactions with WLs are valid, provided that these treatments are randomized within replications.

A line-source irrigation system was used to evaluate trends for DM yield of orchardgrass, perennial ryegrass, intermediate ryegrass, and festulolium cultivars under five WLs (Jensen et al., 2001). Results indicated that production responses across WLs were predominantly linear during summer and fall. It was concluded that DM yield averaged across WLs provided a useful assessment of orchardgrass and ryegrass germplasm for irrigated pastures of the Intermountain Region.

Because WUE as measured by Δ is a potential selection criterion for irrigated pasture grass improvement, an understanding of how Δ is affected by differential water application is important. The present study was conducted to describe (i) the relationship between Δ and DM yield under water limiting conditions in orchardgrass, perennial ryegrass, intermediate ryegrass, and festulolium; and (ii) trends in Δ across four WLs (WL 2, wettest, to WL 5, driest) and two harvest dates under frequent defoliation.

MATERIALS AND METHODS

Nine cultivars each of orchardgrass and ryegrass, which includes intermediate ryegrass cultivar Bison and the festulol-

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Abbreviations: Δ, carbon isotope discrimination; DM, dry matter; PDB, Pee Dee belemnite; WL, water level; WUE, water-use efficiency.

Table 1. Means and trends in carbon-isotope discrimination (Δ) within orchardgrass, perennial ryegrass, intermediate ryegrass, and festulolium cultivars at four water levels, two harvests, combined across 2 yr (1997 and 1998).

Cultivar	Water levels 2	3	4	5	Mean	b†	Linar %‡
	Δ, ‰						
				Orchardgrass			
Ambassador	20.1	19.7	19.6	19.2	19.7	5.23	9.99**
Dawn	19.9	19.4	19.0	18.7	19.3	7.41	97.5**
DS-8	20.0	19.6	19.4	18.9	19.5	6.62	99.1**
Justus	20.1	19.7	19.4	19.1	19.6	6.00	98.3**
Latar (late maturing)	20.1	19.5	19.5	19.1	19.5	5.82	97.2**
Paiute (dryland)	20.3	20.2	19.7	19.3	19.9	6.54	90.2**
Pizza	20.3	19.6	19.5	19.0	19.6	7.64	96.8**
Potomac	20.2	19.7	19.6	19.4	19.7	4.87	94.0*
Sampson	19.6	19.4	19.0	19.0	19.3	4.23	81.4**
LSD (0.05)	0.59	0.52	0.34	0.52	0.40		
				Ryegrass			
Bison-4x§	20.5	19.9	19.6	19.3	19.8	7.16	97.4**
Tandem-4x¶	20.7	20.0	19.8	19.4	20.0	8.02	98.5**
Citadel-4x	20.9	20.3	20.1	20.0	20.3	5.50	89.0**
Bastion-4x	20.8	20.3	19.9	19.8	20.2	6.12	82.5**
Gambit-4x	20.8	20.3	20.0	20.0	20.3	5.84	80.7**
Zero Nui-2x	20.8	20.5	20.3	20.0	20.4	4.87	99.9**
Cambridge-2x	21.4	20.8	20.5	20.3	20.8	7.12	96.7*
Moy-2x	21.3	20.9	20.9	20.3	20.9	5.65	98.3**
Barmaco-2x	21.4	21.3	20.9	20.4	21.0	6.83	87.2**
LSD (0.05)	0.28	0.25	0.32	0.39	0.18		

* Significant at the 0.05 probability level.
** Significant at the 0.01 probability level.
† Regression coefficients of Δ of entries across water levels. Values for water levels (independent variable) expressed as cm of water received by plots.
‡ Percentage of water level sums of squares due to linear effects based on orthogonal polynomials with unequal spacings.
§ Intermediate ryegrass.
¶ Festulolium.

ium cultivar Tandem, were established under a line-source irrigation system with five WLs and subjected to frequent cutting (Table 1). The plant materials used in the study are described in detail by Jensen et al. (2001). All of the ryegrasses were endophyte free. The term dryland refers to plants grown without supplemental irrigation or cultivars selected for performance under nonirrigated conditions. The experiment was located at the Utah State University Evans Experimental Farm, ≈2 km south of Logan, UT (41°45′ N, 111°8′ W, 1350 m above sea level). Soil at the site was a Nibley silty clay loam (fine, mixed, active, mesic Aquic Argixerolls). Ten-year (1990 to 1999) mean annual precipitation at the site was 475 mm, with about half of this amount received from May through October. Total precipitation (excluding irrigation) received from October through September was 584 and 748 mm for 1997 and 1998, respectively. Mean minimum and maximum monthly temperatures in 1997 were 5.6 and 21.8°C for May, 8.7 and 25.4°C for June, 10.8 and 28.7°C for July, 11.4 and 29.7°C for August, 8.4 and 24.5°C for September, and −1.1 and 16.7°C for October. Corresponding values for 1998 were 4.8 and 19.2°C for May, 6.8 and 21.3°C for June, 12.9 and 32.7°C for July, 11.4 and 29.7°C for August, 8.7 and 24.5°C for September, and 0.0 and 16.7°C for October.

Sward plots (16 by 1 m) were planted in early May 1995 with a cone seeder at a depth of 13 mm. The seeding rate for each cultivar was ≈135 seeds per linear meter of row with a 150-mm row spacing. Plots were oriented perpendicular to the irrigation line and divided into five WLs (WL 1 = wettest, WL 5 = driest) on each side of the irrigation line. Each plot was 2.0 m^2 (2 by 1 m) and spaced at 2-m intervals from the line-source sprinkler. The plots were arranged in a modified spilt-plot design with orchardgrass and the ryegrasses as whole plots and the five WLs as subplots. The design was replicated four times, twice on each side of the line-source irrigation line. Samples for carbon-isotope evaluations were taken at WLs 2, 3, 4, and 5 for Harvest 4 (21 July 1997, 18 Aug. 1998) and at WL 2 for Harvest 2 (2 June 1997, 15 June 1998).

Details regarding establishment, fertilizer rates, plot management, and harvest schedules were described by Jensen et al. (2001). Plots were harvested when the forage at WL 5 was between the vegetative and elongation stage (Moore et al., 1991). Mean amounts of water received by the plots for each day from 1 April to Harvest 4 in 1997 were 3.7, 3.0, 2.7, and 2.1 mm d^{-1} for WLs 2 to 5, respectively, and in 1998 were 3.9, 3.2, 2.8, and 2.2 mm d^{-1} for WLs 2 to 5, respectively. Because of above-average precipitation during the spring of 1997 and 1998, a gradient in plant growth was not observed at Harvests 1, 2, and 3. Therefore, sampling across WLs for Δ determinations were delayed until Harvest 4 of each year. Samples for Δ were also taken at WL 2 at Harvest 2 to evaluate the consistency of differences among grass entries at one WL and across two harvests.

Plant samples used for Δ determination were dried at 60°C in a forced-air oven to constant weight, ground in a Wiley mill, and then in a Cyclone mill to pass through a 1-mm screen. Ground plant samples were combusted in a C and N analyzer. The CO_2 and N_2 gases were separated at 50°C on a chromatographic column monitored by a thermal conductivity detector, and each peak was integrated for determination of percentage N and C. The CO_2 gas was then transferred into a trapping system, cryogenically purified, and analyzed for $\delta^{13}C$ [the ratio of $^{13}C/^{12}C$ relative to that of the Pee Dee belemnite (PDB) standard] using an isotope ratioing mass spectrometer. Laboratory precision for $\delta^{13}C$ was better than 0.1‰ (per mil). Standards 21 and 22 (of the National Institute of Standards and Technology) were used routinely to verify values of the working standards. The $\delta^{13}C$ values were converted to Δ as described by Farquhar et al. (1989), assuming that the $\delta^{13}C$ of air on the site was −8‰ on the PDB scale (Mook et al., 1983).

Delta values were analyzed within and across years and

WLs using the GLM procedure (SAS Institute, 1999). A valid F test for the main effect due to WLs was not possible because WLs were not randomized within entries. However, mean squares for entry, and entry × WL were tested with their first-order interactions with replications. Data from individual years were treated as repeated measures in the analysis of data combined across years. Mean separations were made using Fisher's protected LSD at the 0.05 probability level. Linear, quadratic, and cubic trends in Δ across WLs were evaluated using orthogonal polynomials with uneven spacings (Gomez and Gomez, 1984). Amount of water received by the plots (mm d^{-1}) was used in the computation of the coefficients.

RESULTS AND DISCUSSION

Orchardgrass

Harvests 2 and 4, Water Level 2

Samples for Δ were taken at WL 2 from Harvests 2 and 4 to evaluate the consistency of orchardgrass cultivars across harvests. Differences among cultivars for Δ were not significant at Harvest 4 and in the analysis combined across harvests. The cultivars differed significantly ($P < 0.05$, Table 2) for Δ at Harvest 2. There was a significant ($P < 0.05$) harvest × orchardgrass interaction when analyzed within each year, indicating that, in general, orchardgrass cultivars were not consistently ranked for Δ across harvests. A significant correlation between harvests ($r = 0.46$, $P = 0.05$) and the lack of variation for Δ within orchardgrass cultivars, suggests that the interactions had little biological significance. The dryland orchardgrass cultivar Paiute had the highest Δ values at Harvests 2 and 4, while 'Sampson' had the lowest Δ values at both harvests.

The overall trend from Harvests 2 to 4 was for increased Δ. Combined across years, mean Δ value for Harvest 2 was 19.9‰ compared with 20.1‰ for Harvest 4. With increased average temperatures from Harvests 2 to 4 and reduced forage yields (Jensen et al., 2001), it was expected that Δ values should have declined (McCree and Richardson, 1987; Hall et al., 1994). The observed increase in Δ from Harvests 2 to 4 may be attributable to elongated stems (Moore et al., 1991) in the samples for Harvest 2, as compared with Harvest 4 samples from completely vegetative plants. Johnson and Asay (1994–1995, unpublished data) found that Δ values for stems of crested wheatgrass were 16.5‰ compared with a Δ of 18.5‰ for flag leaves.

Table 2. Mean squares from analysis of variance for carbon isotope discrimination (Δ) from orchardgrass, perennial ryegrass, intermediate ryegrass, and festulolium cultivars at Water Level 2 across two harvest dates and two years (1997 and 1998).

Source of variation	df	Harvest 2†	Harvest 4‡	Combined
		Δ, ‰		
		Orchardgrass		
Cultivar (C)	8	0.2934*	0.1622	0.4148
Harvest (H)	1	–	–	0.7368
C × H	8	–	–	0.0409
Year (Y)	1	4.1391**	0.2885*	3.3065*
C × Y	8	0.0034	0.0337	0.1716
H × Y	1	–	–	1.1211
C × H × Y	8	–	–	0.2387**
		Ryegrass		
Cultivar (C)	8	0.1425	0.4097**	0.4316**
Harvest (H)	1	–	–	0.4150*
C × H	8	–	–	0.1206*
Year (Y)	1	1.1333	0.9629**	2.0928*
C × Y	8	0.0722	0.2454**	0.1561
H × Y	1	–	–	0.0035
C × H × Y	8	–	–	0.1665*

* Significant at the 0.05 probability level.
** Significant at the 0.01 probability level.
† Harvest 2, 2 June 1997 and 15 June 1998.
‡ Harvest 4, 21 July 1997 and 18 Aug. 1998.

Harvest 4, Water Levels 2 to 5

Variation for Δ appears to be limited in the orchardgrass cultivars studied. Combined across years and within WLs, differences among cultivars for Δ at Harvest 4 were not significant except for WL 4 (Table 3). However, analysis of variance within years and WLs indicated that Δ varied significantly ($P < 0.05$) among orchardgrass cultivars for WLs 2 and 5 in 1997 and for WLs 3 and 4 in 1998. Delta ranged from 19.2‰ for Dawn to 19.7‰ for Paiute across WLs in 1997, and from 19.2‰ for Sampson to 20.1‰ for Paiute in 1998. The dryland cultivar, Paiute, had the highest Δ values when compared with the mean of the irrigated (i.e., cultivars developed under optimum irrigation) orchardgrass cultivars at all WLs (Fig. 1). A significant ($P < 0.05$) year × orchardgrass interaction was attributed to decreased Δ at WL 2 for Paiute and increased Δ in the irrigated orchardgrass cultivars at WL 2 in 1997.

In general, Δ decreased as water stress increased. Averaged across cultivars and years, Δ was 20.1‰ for WL 2, 19.6‰ for WL 3, 19.4‰ for WL 4, and 19.1‰ for WL 5. Relative differences in Δ among orchardgrass cultivars were consistent across WLs in both 1997 and 1998, as evidenced by a nonsignificant orchardgrass × WL interaction (Table 3). In general, the irrigated cultivars of orchardgrass had lower Δ values than did the dryland cultivar Paiute. Perhaps most noteworthy is that DM yield for Paiute was equivalent to or significantly greater than all other entries at WLs 4 and 5 (Jensen

Table 3. Mean squares from analysis of variance for carbon isotope discrimination (Δ) from orchardgrass, perennial ryegrass, intermediate ryegrass, and festulolium cultivars at Harvest 4 within water levels across 2 yr (1997 and 1998).

Source of variation	df	Water level 2	3	4	5	Combined
		Δ, ‰				
		Orchardgrass				
Cultivar (C)	8	0.1622	0.2187	0.2478*	0.1572	0.6073
C × WL	24	–	–	–	–	0.0595
Year (Y)	1	0.2885	0.0515	0.0231	1.1817**	0.6000*
C × Y	8	0.1828	0.0689*	0.0391	0.0208	0.1356*
WL × Y	3	–	–	–	–	0.3149
C × WL × Y	24	–	–	–	–	0.0587
		Ryegrass				
Cultivar (C)	8	0.4097**	0.8651**	0.8563**	0.5506**	2.1337**
C × WL	24	–	–	–	–	0.0736*
Year (Y)	1	0.9629**	0.0703	0.2956	1.9645*	0.8119*
C × Y	8	0.2454**	0.1609**	0.0682	0.0675	0.3023**
WL × Y	3	–	–	–	–	0.9253**
C × WL × Y	24	–	–	–	–	0.0764*

* Significant at the 0.05 probability level.
** Significant at the 0.01 probability level.

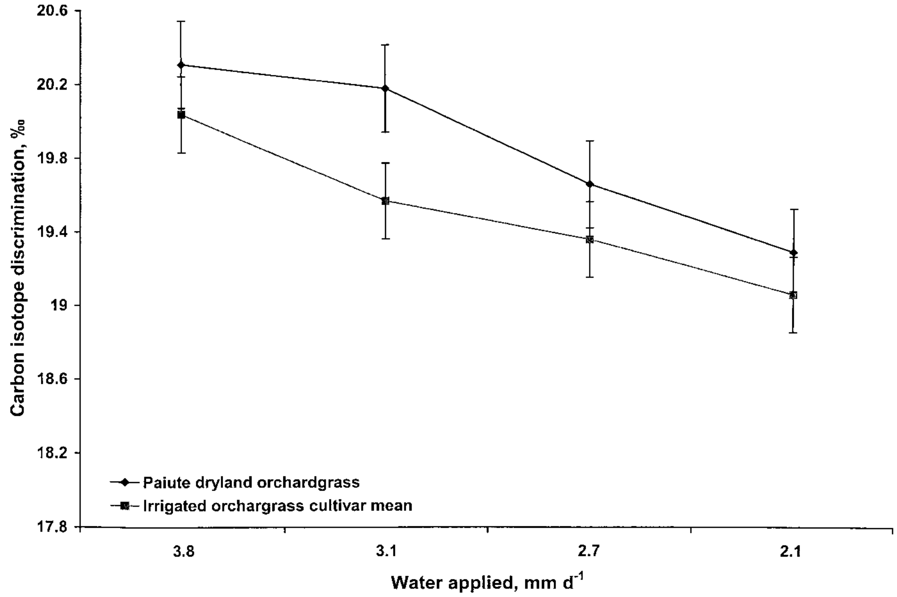

Fig. 1. Trends in carbon isotope discrimination (Δ) across four water levels for the dryland orchardgrass cultivar Paiute and the mean of the irrigated orchardgrass cultivars across 2 yr (1997 to 1998) at the Evans Research Farm near Logan, UT. Bar = SE.

et al., 2001). This is contrary to what would be expected based only on WUE (Johnson and Tieszen, 1994). However, other factors also influence production under limiting water, including more extensive root exploration for soil water or more effective water extraction characteristics, thereby maintaining a better water balance and greater DM yield with less irrigation. Within WLs, Δ did not differ significantly between early and late maturing orchardgrass cultivars.

With the exception of Potomac, the sums of squares due to linear trends in Δ across WLs was significant ($P < 0.05$) for each year and in the analysis combined across years (Table 1). Linear trends for Potomac were significant ($P < 0.05$) in 1997. In 1998 there was an increase in Δ from WLs 2 to 3 with no further declines at WLs 4 and 5. Quadratic and cubic trends for Δ were not significant across WLs within years and across years (Table 1). Correlations between DM yield and Δ were not significant for WL 2 to 4 in orchardgrass (Table 4).

Table 4. Pearson correlation coefficients (*r*) between dry matter yield (DMY) and carbon isotope discrimination (Δ) with four water levels (WL) form a line-source irrigation system combined across years (1997 to 1998).

	Δ, ‰			
DMY	WL 2	WL 3	WL 4	WL 5
Mg ha^{-1}				
		Orchardgrass		
WL 2	−0.44ns†	−0.21ns	−0.33ns	−0.16ns
WL 3	−0.41ns	−0.45ns	−0.36ns	−0.24ns
WL 4	−0.53ns	−0.63ns	−0.53ns	−0.44ns
WL 5	−0.57ns	−0.69*	−0.64ns	−0.68*
		Ryegrass		
WL 2	−0.31ns	−0.36ns	0.77*	0.85**
WL 3	−0.24ns	−0.33ns	0.75*	0.86**
WL 4	−0.42ns	−0.46ns	0.69*	0.79*
WL 5	−0.51ns	−0.52ns	0.75*	0.80**

* Significant at the 0.05 probability level.
** Significant at the 0.01 probability level.
† ns, not significant.

With increased drought, the negative association between DM yield and Δ became significant ($P < 0.05$) at WL 3 ($r = -0.69$) and WL 5 ($r = -0.68$), suggesting that selection for genotypes with increased DM yield under drought would likely decrease Δ.

Ryegrasses

Harvests 2 and 4, Water Level 2

Comparative differences among ryegrasses for Δ were not consistent across harvest dates, as indicated by a significant ($P < 0.05$) ryegrass by harvest interaction. A significant correlation between harvests ($r = 0.46$, $P = 0.05$) indicated that although some differences among cultivars were inconsistent across harvest dates, others were consistent. This interaction was due to a large increase in Δ from Harvests 2 to 4 for cultivars Cambridge and Moy, and several ranking changes between harvests for other cultivars. Even though not significant, diploid ($2n = 2x = 14$) perennial ryegrass tended to have higher Δ values than tetraploid ($2n = 4x = 28$) ryegrasses at both harvests. Mean Δ values for Harvest 2 were 20.9‰ and 20.7‰ compared with Harvest 4 Δ values of 21.2‰ and 20.7‰ for the diploids and tetraploids, respectively.

Differences among ryegrass cultivars for Δ were not significant at Harvest 2. The cultivars differed significantly ($P < 0.01$) for Δ at Harvest 4 and in the analysis combined across harvests ($P < 0.01$, Table 2). Mean Δ values ranged from 20.6‰ for Bastion to 21.2‰ for Barmaco at Harvest 2 and from 20.5‰ for Bison to 21.4‰ for Cambridge at Harvest 4.

Harvest-4, WLs 2 to 5

Significant ($P < 0.01$) differences in Δ values were found among ryegrass cultivars within each WL (Table 3). Values of Δ ranged from 21.3‰ for Barmaco to 19.0‰ for Tandem across WLs in 1997, and from 21.8‰ for

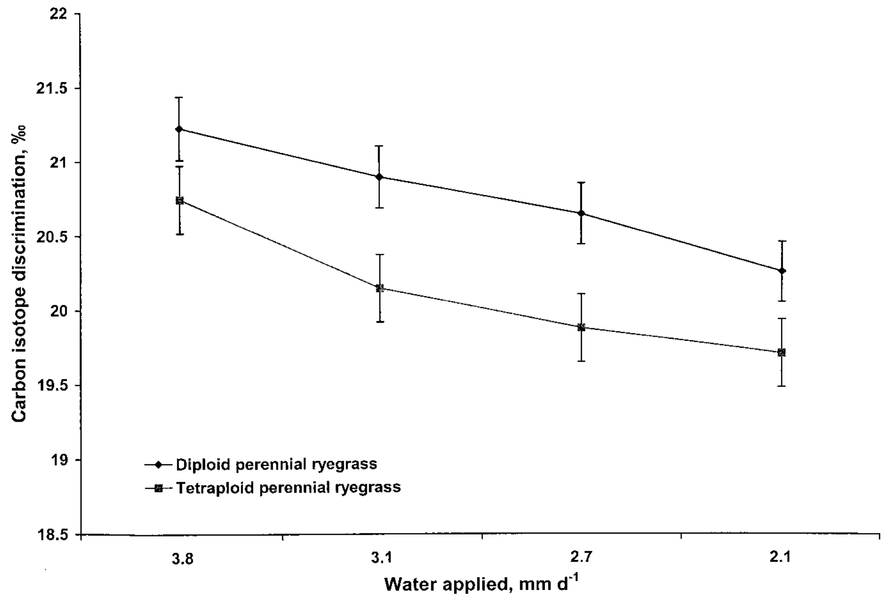

Fig. 2. Trends in carbon isotope discrimination (Δ) across four water levels for diploid and tetraploid ryegrass cultivars across 2 yr (1997 to 1998) at the Evans Research Farm near Logan, UT. Bar = SE.

Barmaco to 19.2‰ for Bison in 1998. Relative differences in Δ among ryegrass cultivars were not consistent across WLs in 1997 and the combined analysis as evidenced by a significant ($P < 0.05$) ryegrass × WL interaction. This interaction was attributed to an increase in Δ values at WLs 3 and 4 in 1997 for Barmaco and Moy, while Δ declined for all other cultivars with increased drought.

Combined across years and within WLs, tetraploid ryegrass cultivars had significantly lower Δ with a mean of 20.1‰ compared with 20.8‰ for the diploids (Fig. 2). Jensen et al. (2001) reported that tetraploid ryegrass cultivars produced more DM at corresponding WLs than did diploid cultivars. Similar trends in Δ values were reported between diploid and tetraploid Russian wildrye [*Psathyrostachys juncea* (Fisch.) Nevski] entries (Asay et al., 1996).

With the exception of Citadel and Bastion, which had significant linear and quadratic trends with increased drought, all other ryegrass cultivars had a linear ($P <$ 0.05) decline in Δ as water became limiting (Table 1). Cultivars Zero_nui and Citadel were the most stable across WLs, as evidenced by their low b-values (Table 1). Ryegrass cultivars Tandem and Bison were the most responsive for Δ with increased drought.

At the higher WLs, correlations between Δ and DM yield were negative, but not significant (Table 4), while at WLs 4 and 5 the correlations became positive and significant ($P < 0.05$). The largest significant ($P < 0.01$) correlations between Δ and DM yield occurred at WL-2 ($r = 0.86$), WL 3 ($r = 0.85$), and WL 5 ($r = 0.80$).

The use of Δ as a selection criterion and the corresponding influence on DM yield is still a matter of uncertainty. Johnson and Bassett (1991) reported a significant negative correlation between DM yield and Δ under both irrigated and dryland conditions for orchardgrass and ryegrasses in southeastern Washington. However, in the same study, no association between Δ and DM yield was observed in eastern Washington. Variable relationships between Δ and DM yield also have been reported in crested wheatgrass (Johnson et al., 1990; Read et al., 1991, 1993; and Asay et al., 1998) and wheat (Condon et al., 1987).

In summary, within all WLs, orchardgrass cultivars expressed significantly ($P < 0.05$) lower Δ than all ryegrass cultivars. More variation for Δ was observed in ryegrass cultivars than orchardgrass cultivars. The lack of variation in orchardgrass may be a reflection of the narrow genetic base within the orchardgrass cultivars compared with a much broader genetic base within the ryegrass cultivars. Our results indicated that both species can be effectively evaluated for Δ across irrigation levels and harvest dates. Negative correlations between Δ and DM yield for orchardgrass (Table 4) suggests that selection for increased WUE would likely result in increased DM yield. However, under water stress (WLs 4 and 5), positive correlations between Δ and DM yield for the ryegrasses suggests that selection for improved WUE would lead to decreased DM yield. To avoid possible declines in DM yield in breeding programs to decrease Δ in orchardgrass and ryegrasses, DM yield also should be monitored.

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